

Amendments to the Claims

Claim 1 (original): A method for reducing total sample complexity in native or digested biological sample(s), before analysis thereof by mass spectrometry, comprising the following steps:

- a) selecting a fraction from the entire native or digested biological sample(s) on the basis of pI-value, said fraction comprising native or digested sample representing the entire substance population in the sample;
- b) separating native or digested sample substances from each other; and
- c) analysing said substances by mass spectrometry.

Claim 2 (currently amended): ~~A method according to~~ The method of claim 1, wherein said substances are peptides obtained from proteins in the sample(s).

Claim 3 (currently amended): ~~A method according to~~ The method of claim 1-~~or~~2, wherein the pI-value is 3.5 - 4.5 or a sub range thereof.

Claim 4 (currently amended): ~~A method according to~~ The method of claim 1-~~or~~2, wherein the pI-value is selected to target one or more specific peptides.

Claim 5 (currently amended): ~~A method according to~~ one or more of the above claims, The method of claim 1, wherein said fraction in step a) is obtained by anion exchange chromatography.

Claim 6 (currently amended): ~~A method according to~~ The method of claim 5, wherein the separation in step b) is by cation exchange chromatography.

Claim 7 (currently amended): ~~A method according to one or more of the above claims,~~ The method of claim 1, wherein, in step a), the sample is dissolved in a buffer with pH 4.5, the sample is loaded onto an anion exchange column, and the desired peptides are eluted in a buffer with pH 3.5.

Claim 8 (currently amended): ~~A method according to one or more of the above claims,~~ The method of claim 1, wherein the separation in step b) is by multidimensional chromatography, MDLC, comprising cation exchange chromatography, RPC (reverse phase chromatography) and MS/MS.

Claim 9 (currently amended): ~~A method according to one or more of the above claims,~~ The method of claim 6, wherein the anion exchange column is coupled to the cation exchange column.

Claim 10 (currently amended): ~~A method according to claims 8 or 9,~~ The method of claim 9, wherein the pH in step a) is higher than in step b).

Claim 11 (currently amended): ~~A method according to any of the claims 1-4,~~ The method of claim 1, wherein the fraction in step a) is obtained by isoelectric focussing.

Claim 12 (currently amended): ~~A method according to any of the claims 1-4,~~ The method of claim 1, wherein the fraction in step a) is obtained by chromatofocussing.

Claim 13 (currently amended): ~~A method according to The method of claim 11 or 12, which is integrated to a conventional MDLC (multidimensional liquid chromatography) flow path.~~

Claim 14 (currently amended): ~~A method according to one or more of the above claims, The method of claim 1, wherein the mass spectrometric analysis is tandem MS.~~

Claim 15 (currently amended): ~~A method according to one or more of the above claims, The method of claim 1, wherein the MS is ESI (electrospray ionisation)-MS.~~

Claim 16 (currently amended): ~~A method according to one or more of the claims 1-14, The method of claim 1, wherein the MS is MALDI (matrix assisted laser desorption ionisation)-MS.~~

Claim 17 (currently amended): ~~A method according to one or more of the above claims, The method of claim 1, wherein the biological sample(s) comprises at least two samples which are differentially labelled.~~

Claim 18 (currently amended): A system for reducing total sample complexity ~~in a method according to one or more of the claims 1-17, comprising a charge-selective column coupled to a MDLC work flow path comprising a cation exchange column and a RPC column.~~

Claim 19 (currently amended): ~~A system according to~~ The system of claim 18, wherein the charge-selective column is an anion exchange column.

Claim 20 (currently amended): ~~A system according to~~ The system of claim 18 or 19, wherein the charge-selective column is run with a first buffer having pH 4.5-4.0 and a second buffer having pH 3.5-4.0, wherein the second buffer has lower pH than the first buffer and is used for elution.

Claim 21 (currently amended): ~~A system according to~~ The system of claim 18, wherein the charge-selective column is a chromatofocussing column.

Claim 22 (currently amended): ~~A system according to~~ The system of claim 18, wherein the charge-selective column is an isoelectric focussing column.

Claim 23 (currently amended): ~~A system according to one or more of the claims 20-22, The system of claim 20,~~ wherein the cation exchange column is run with a third buffer with pH lower than the buffer used for elution from the charge-selective column.

Claim 24 (new): The method of claim 12, which is integrated to a conventional MDLC (multidimensional liquid chromatography) flow path.